correlated but disassembly occurs more readily. This indicates monomers form faster than the quaternary structures. Characteristics of monomer-monomer interfaces were observed between 1YEP chain A and chain B and BCP1 (3DRN) while the characteristic of dimer-dimer interface was seen between 1YEP chain B and chain C.

3408-Pos Board B136

Investigating Characteristics of Folding Cores from Analyses of Folding Mechaniams on Multi-Transition Proteins by Means of a Coarse-Grained Go Model

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Though it has become clear that small and fast folding proteins have funnel shaped energy landscape, we have not know explicitly what and when happens in folding process. The knowledge of precise picture in each transition in the folding process of multi state proteins should provide the hints to elucidate the detailed folding processes.

However, many hypotheses that try to explain the mechanisms of multi-state folding have been proposed (such as hypotheses regarding the packing of side chains after searching the protein's topology, those regarding a number of subdomains that form a distinct barrier, those regarding discrete two-dimensional and three-dimensional structure formation, and others) and we have not yet gained a precise understanding.

To understand the mechanisms of multi-state folding, thermodynamic contributions of each component of the system, such as a main-chain, a side-chain and water, should be specified. It can be accomplished by comparing the simulation results of some models which has different resolution such as Ca-Go model, full-atom Go model and all-atom with explicit water.

In the precedent study, as a first step, we showed that the difference in the shape of the free energy profiles could be interpreted by the number of the regions that folds cooperatively and degree of coupling of the regions by means of modified Ca-Go model.

In this study, we apply the same method with previous work to other proteins which are suggested to fold into the native structures through multiple transitions and affirm the number of the barrier and stability of the intermediate states. We also investigate that the characteristics of the regions to contribute to barrier constructions.

3409-Pos Board B137

Effects of Energetic Heterogeneity on Protein Folding Dynamics Across Many Non-Homologous Proteins

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Native structure based models (SBMs, ''Go-models'') are based on energy landscape theory and the principle of minimal frustration [1,2]. Using this framework strongly reduces computational demands and allows us to investigate the influence of the contact-energy weight onto the folding process for many different protein families. A large set (approx. 200) of non-homologous monomeric proteins sized from 50-150 amino acids are simulated on a coarse-grained level, representing each amino acid by a single bead. A fully automatized workflow implemented with the help of eSBMTools [3] allows to quantify typical folding properties like phi-values, folding free energy landscapes and transition state ensembles (TSEs) for the simulated proteins.

Conventional SBMs use sequence independent homogeneous energy weights for the native contact energy. We compare these ''vanilla'' models with sequence dependent heterogeneous ''flavored'' energy weights as introduced by Miyazawa and Jernigan [4]. Subsequently, the dynamics of forming TSEs are compared by the sequential formation of tertiary interfaces between secondary structure elements in vanilla and flavored simulations. We find that, despite the energetic dissimilarity, the sequential ordering is similar between both types of simulations and appears as a robust property. This suggests that protein folding dynamics are strongly influenced by the native protein topology.

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3410-Pos Board B138

Aromatic Amino Acids Promote Peptide Folding by Reducing Backbone Hydration

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The relation between the sequence of a protein and its tridimensional structure remains largely unknown. To better understand how single residues impact on the local protein structure, we studied peptides of sequence EGAAXAASS (X = Gly, Ile, Tyr, Trp) through comparison of molecular dynamics (MD) trajectories and NMR residual dipolar coupling (RDC) measurements. The RDC patterns of the peptide with X = Gly or Ile are rather flat, suggesting extended, unfolded peptides, while the contrasted patterns for peptides with X = Tyr or Trp suggest compact folded structures. The comparison shows that the formation of internal hydrogen bonds underlying helical-turns is key to reproduce experimental RDC values for the peptides containing aromatic residues. The simulations further reveal that the driving force leading to such helical-turn conformation arises from the lack of hydration of the peptide chain on either side of the bulky aromatic side chain, which can potentially act as a nucleation point initiating the folding process. These results provide a starting point to understand the amino acid code underlying the mechanism of protein folding.

3411-Pos Board B139

pH Dependent Conformational Change of Hepcidin and its Precursor Protein, Pro-Hepcidin

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Hepcidin plays a role in the regulation of iron homeostasis through its interaction with an iron transpoter protein, ferroportin, in the liver. Hepcidin consists of 25 amino acid residues and four intra-molecular disulfide bonds, which are absolutely required for its biological activity, not only for iron homeostasis, but also for anti-microbial activity. To investigate structure-function relationships, hepcidin was chemically synthesized. However, it is well known that the synthetic yield of hepcidin is quite low under the typical folding conditions. To overcome this issue and regulate the disulfide-coupled folding of hepcidin, we studied the conformation of reduced/denatured hepcidin by means of CD spectroscopy under a variety of conditions. In addition, recombinant pro-hepcidin was also prepared using *E. coli* expression system and its conformation investigated, since the disulfide-coupled folding of hepcidine proceeds via a precursor *in vivo*. The major problem of the disulfide-coupled folding of hepcidin is that it undergoes aggregation during its folding reaction. To solve this problem, several types of redox reagents and solvents were examined to improve the folding efficiency of hepcidin. However, all of the reagents resulted in quite low yields for the disulfide-coupled folding of hepcidin. Therefore, we estimated the pH of the folding solution used to regulate disulfide-coupled folding of hepcidin since the folding velocity and conformation of disulfide-containing peptide is significantly affected by the pH of the solution. Our experimental results show that reduced/denatured hepcidin is not stable around neutral pH's, although the native hepcidin is quite soluble in aqueous buffers at a neutral pH. Alkaline conditions were also not effective for the folding reaction. Therefore, we studied the conformation of hepcidin and pro-hepcidin under acidic conditions. The results will be discussed in this paper.

3412-Pos Board B140

Myosin UNC-45 Chaperone: The Role of its Domains in the Interaction with the Myosin Motor Domain

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The proper folding of many proteins can only be achieved by the interaction with molecular chaperones. The molecular chaperone UNC-45 is required for the folding of striated muscle myosin II, however, the precise mechanism by which it contributes to proper folding remains unclear. UNC-45 contains three domains: an N-terminal TPR domain known to bind to Hsp90, a central domain of unknown function, and a C-terminal UCS domain known to interact with the myosin head. Here we used fluorescence titrations methods, dynamic light scattering, and single-molecule atomic force microscopy (AFM) unfolding/refolding techniques to study the interactions of the UCS and central domains with the myosin motor domain. To study the effects of UNC-45, UCS and central domain binding to the myosin motor domain we used environmentally sensitive fluorescence as a reporter for the UNC-45 domain-myosin interaction. We found that the UCS domain and the central domain bind to distinct sites on the myosin motor domain. In order to test which UNC-45 domain has a chaperone activity we used two different methods: i) prevention of misfolding using single-molecule AFM, and ii) prevention of aggregation using dynamic light scattering manner. Using the first method we found that the UCS domain is sufficient to prevent misfolding of a titin mechanical